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Extractive spectrophotometric determination of levetiracetam in pharmaceutical formulations

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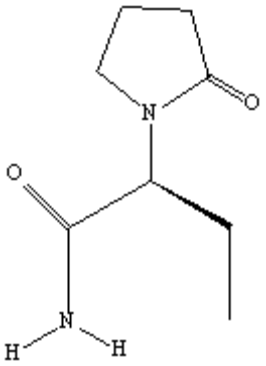
ABSTRACT

Three simple, economical, precise, reliable and reproducible visible spectrophotometric methods (A, B and C) have been developed for the estimation of Levetiracetam in bulk as well as in Tablet formulation. The developed methods A, B and C are based on the formation of chloroform extractable complex of Levetiracetam with Bromocresol green (method A), Bromophenol blue (method B) and Bromothymol blue (method C) which shows absorbance maxima at 435 nm, 454 nm and 415 nm respectively. The absorbance-concentration plot is linear over the range of 2.5-25 mcg/ml for Method A, 2.5-25mcg/ml for Method B and 1.5-15 mcg/ml for Method C. The different experimental parameters affecting the development and stability were studied carefully and optimized. Results of analysis for all the methods were validated statistically and by recovery studies.

Keywords: Levetiracetam, Bromocresol green, Bromophenol blue, UV double beam spectrophotometer.

INTRODUCTION

Levetiracetam [1] is a novel antiepileptic agent; with a chemical name (S)-(2)-(2-oxopyrrolidin-yl) butamide with a molecular formula $C_8H_{14}N_2O_2$ and a molecular weight of 170.20. It is used as an adjunctive therapy in the treatment of partial seizures[2]. Literature survey reveals many Chromatographic methods[3-8] for the determination of Levetiracetam, in biological fluids and Formulations. Therefore the need for fast, low cost and selective method is obvious especially for routine Quality Control analysis of pharmaceutical formulation.



EXPERIMENTAL

Instrument

Shimadzu double beam Ultraviolet-Visible double beam spectrophotometer with 1 cm matched quartz cells was used for all spectral measurements.

Reagents

All the chemicals used were of analytical reagent grade. Acid phthalte buffer pH 2.4 and pH 3 were prepared as per I.P.

- Bromocresol green, BCG (0.1% w/v)-100 mg is weighed accurately and dissolved in 100 ml of acid phthalate buffer pH 2.4.
- Bromophenol Blue, BPB (0.1% w/v)- 100 mg is weighed accurately and dissolved in 100 ml of acid phthalate buffer pH 3.0.
- Bromothymol Blue, BTB (0.1% w/v)-100 mg is weighed accurately and dissolved in 100 ml of acid phthalate buffer pH 2.4.
- Chloroform AR grade

Procedure

Standard stock solution: A standard stock solution containing 1mg/ml was prepared by dissolving 100 mg of Levetiracetam in 100 ml of water. From this, a working standard solution containing 50mcg/ml was prepared with water.

Assay procedure

Method A

Aliquots of the drug solution of Levetiracetam 0.5-5.0 ml (50mcg/ml) are taken and transferred into a series of 125 ml of separating funnel. To each separating funnel 2 ml of BCG reagent solution is added. Reaction mixture was shaken gently for 5 min. Then 10 ml of chloroform was added to each of them. The contents are shaken thoroughly for 5 min and allowed to stand, so as to separate the aqueous and chloroform layer. Colored chloroform layer was separated out and absorbance was measured at 435 nm against reagent blank. Calibration curve was prepared from absorbance values so obtained

Method B

Aliquots of the drug solution of Levetiracetam 0.5-5.0 ml (50mcg/ml) are taken and transferred into a series of 125 ml of separating funnel. To each funnel 5 ml of BPB reagent is added. Reaction mixture was shaken gently for 5 min. Then 10 ml of chloroform was added to each of them. The contents are shaken thoroughly for 5 min and allowed to stand, so as to separate the aqueous and chloroform layer. Colored chloroform layer was separated out and absorbance was measured at 454 nm against reagent blank. Calibration curve was prepared from absorbance values so obtained.

Method C

Aliquots of the drug solution of Levetiracetam 0.3-3.0 ml (50mcg/ml) are taken and transferred into a series of 125 ml of separating funnel. To each funnel 5 ml of BTB reagent is added. Reaction mixture was shaken gently for 5 min. Then 10 ml of chloroform was added to each of them. The contents are shaken thoroughly for 5 min and allowed to stand, so as to separate the aqueous and chloroform layer. Colored chloroform layer was separated out and absorbance was measured at 415 nm against reagent blank. Calibration curve was prepared from absorbance values so obtained.

Preparation of sample solution

Tablets containing Levetiracetam were successfully analyzed by the proposed methods. Twenty tablets of Levetiracetam (Levroxa 250mg, Ranbaxy) were accurately weighed and powdered. Tablet powder equivalent to 100 mg of Levetiracetam was dissolved in 50 ml of water and filtered and washed with water, the filtrate and washings were combined and the final volume was made to 100 ml with water. The solution was suitably diluted and analyzed as given under the assay procedure for bulk samples. The results are represented in Table 2. None of the excipients usually employed in the formulation of tablets interfered in the analysis of Levetiracetam, by the proposed methods.

Recovery Studies: To ensure the accuracy and reproducibility of the results obtained, adding known amounts of pure drug to the previously analysed formulated samples and these samples were reanalyzed by the proposed method performed recovery experiments. The percentage recoveries thus obtained were given in Table 2.

RESULTS AND DISCUSSIONS:

In the present work three methods have been developed for the estimation of Levetiracetam

Table 1: Optical characteristics and precision data

	Method A	Method B	Method C
λ_{max} (nm)	435	454	415
Beer's law limits (mcg/ml)	2.5-25	2.5-25	1.5-15
Molar absorptivity (l/mol.cm)	$2.42x10^3$	$2.63x10^3$	$3.80x10^3$
Sand ell's sensitivity(micrograms/cm^2/0.001 absorbance unit)	0.24272	0.22272	0.15432
Regression Equation* (Y)			
Slope (m)	0.004127	0.004465	0.00657
Intercept (c)	0.007529	0.267956	0.58929
Correlation Coefficient(r)	0.9999	0.9998	0.9998
Precision (%Relative Standard Deviation)	0.9499	0.7952	0.5643
Intra Precision (%Relative Standard Deviation)	0.6495	0.8285	0.7964
Standard error of mean	0.0039	0.0035	0.0037

*Y=mx+c, where X is the concentration in micrograms/ml and Y is absorbance unit.

Table 2: Assay of levetiracetam in tablets

Sample No.	Labelled Amount (mg)	% Obtained* by proposed method			**% Recovery by the Proposed method		
		MethodA.	MethodB	MethodC	Method A	Method B	Method C
1	250	99.8	99.9	100.3	100.1	99.9	100.2
2	250	100.2	100.2	99.9	100.2	100.3	100.5

*Average of three determinations.

** After spiking the sample.

from Tablet formulation. The developed Methods A, B and C are based on formation of chloroform extractable yellow colored ion-pair complexes with BCG, BPB and BTB respectively. The conditions required for the formation of colored complexes were optimized. Statistical analysis was carried out and the results of which were satisfactory. Relative Standard Deviation values were low that indicates the reproducibility of the proposed methods. Recovery studies were close to 100 % that indicates the accuracy and precision of the proposed methods.

The optical characteristics such as absorption maxima, Beer's law limits, molar absorptivity and Sand ell's sensitivity are presented in Table I.

The regression analysis using the method of least squares was made for slope (m), intercept (b) and correlation obtained from different concentrations and the results are summarized in Table 1.

In conclusion, the proposed methods are simple, economical, sensitive, precise reliable and reproducible for the routine estimation of Levetiracetam in bulk as well as in tablet formulation.

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